For the agenda for each of the two topics, background statements from relevant documents are quoted, followed by several points provided as a starting basis for discussion.

Agenda: Bioaccumulation

The original direction to Teck regarding the Problem Formulation work plan included the following (which was drafted to be consistent with the Sediment LOE):

"Invertebrate-tissue chemistry represents an important line of evidence for evaluating risks to benthic invertebrates associated with exposure to COIs in the study area (see Sediment LOE, Section 9). More specifically, data from laboratory bioaccumulation tests provide information on the bioavailability of sediment-associated COIs and on their accumulation in invertebrate tissues. Matching tissue chemistry and sediment toxicity data from laboratory toxicity tests can provide the information needed to identify critical body residues of COIs. In turn, this information can be used to interpret field collected invertebrate-tissue chemistry data. Two laboratory-based and one field method for evaluating bioaccumulation and tissue resides at the UCR are recommended.

Lab-based bioaccumulation tests may be used to evaluate the relationships between invertebrate tissues and toxicity in benthic organisms. Synoptic bioaccumulation testing with sediment toxicity testing and chemical analyses would support the interpretation of sediment toxicity studies, provide information for estimating dietary exposure to COIs, estimating bioaccumulation functions (i.e., BSAFs, BCFs), and may support the development of relationships between invertebrate-tissue chemistry and the responses of benthic invertebrates. These latter relationships may be used to evaluate the invertebrate-tissue chemistry data collected in the field. The following tests are requested or potentially useful:

- Tissue residues (i.e., body burdens) should be measured in midge larvae exposed to UCR sediment under controlled laboratory conditions. More specifically, midge for analysis of tissue chemistry should be obtained from additional replicates of selected samples (e.g., 15 to 20 samples [not including reference samples]) used for the 10-d whole-sediment toxicity tests (e.g., exposure for tissue chemistry will be conducted concurrently with the toxicity tests).
- 28-d whole-sediment bioaccumulation tests with the oligochaete, *Lumbriculus* variegatus, should be conducted using splits of the same 15 to 20 sediment samples selected for analysis of midge tissues. Test methods should follow appropriate guidance (e.g., USEPA 2000 and ASTM 2011d). Sediment samples for bioaccumulation testing should be selected to provide a large gradient of metal concentrations in sediment.
- Tissue concentrations in field collected organisms are a potential third method for
 evaluating exposure of COPCs to benthic macroinvertebrates at the site. Paired
 sediment chemistry data with tissue concentrations could also inform several of the
 objectives listed above, but are not necessarily required for the BERA, nor helpful for
 interpreting the results of laboratory bioassays."

The draft Problem Formulation Work Plan submitted by Teck did not include any of the three bulleted items, but proposed this analysis plan, with no laboratory bioaccumulation tests:

"Information on benthic invertebrate tissue concentrations are used as inputs into fish and wildlife dietary models. High fish tissue concentrations may be representative of areas where benthic invertebrate tissue concentrations would be high. Fish tissue could represent an average across invertebrate types and locations, depending upon fish foraging preferences. Therefore, the expected maximum or average amount of chemical in fish tissue may be used as a first approximation of the upper bound of chemical concentrations in benthic invertebrates (i.e., none of the COPCs are known to have significant biodilution between the invertebrate and vertebrate trophic levels). Locations with high sediment concentrations of bioavailable COPCs and high fish tissue residues (based on small to medium sized insectivorous fish with relatively small home ranges) might represent areas that could be sampled, whereas areas with very low sediment concentrations or fish tissue concentrations below dietary thresholds might not need to be sampled. Selection of locations for collection of benthic invertebrates also should be representative of all aquatic habitat types (riverine, transitional, lacustrine, wetland), applicable sediment gradients (see Aquatic Invertebrate Communities section), and wildlife feeding areas (wetlands). Synoptically collected sediment samples might be collected to verify that habitat types and gradients were adequately represented."

As well, the draft Sediment Benthic Toxicity QAPP submitted by Teck does not contain these tests for bioaccumulation.

DOI says (April 2011; comments on draft Sediment QAPP):

"First, the midge required for analysis of tissue chemistry should be obtained from additional replicates of 15 to 20 sediment samples used to conduct 10-d toxicity tests with midge, *C. dilutus*. In addition, 28-d whole-sediment bioaccumulation tests with oligochaetes (*L. variegatus*) should be conducted using splits of 15 to 20 of the same sediment samples that will be used to conduct the 10-d toxicity tests with midge. These samples were to be selected to obtain a large gradient in sediment metal concentrations. This study was to be conducted to evaluate the bioavailability of sediment-associated COPCs in the UCR and to support identification of critical body residues of COPCs in invertebrate tissues. To supplement the data collected in the laboratory, invertebrate-tissue samples (two or three taxa per location) were to be collected from 25 or more locations within the riverine portion of the UCR."

EPA says (Sept. 14, 2011; letter to PPs):

"We are not planning to require separate bioaccumulation tests since we will have more direct information on bioaccumulation from tissue analyses. Our current thinking is that we don't need a separate test to measure contaminant bioaccumulation from sediment up to benthic invertebrates because we won't be modeling up from sediment to fish using a food chain model. Instead, we will use the directly measured concentrations of contaminants in fish and other prey (mussels, possibly crayfish) in dietary models for other fish, birds, and mammals. We do plan to

ask Teck to retain the animals from the 28 day *Hyalella azteca* chambers, and to run extra chambers if necessary to generate sufficient tissue mass for chemical analysis, which could then be used to evaluate bioaccumulation. As discussed during our April, 2011 meeting in Seattle, measurement of residues in *Hyalella* could serve the same general purpose as the previously discussed assays with *Lumbriculus*, but would have the advantage of building on published residue-response information, which is more advanced for *Hyalella* than for *Lumbriculus*. The purpose of this work would be to support the interpretation of the bioassay data. Bioaccumulation is another measure (in addition to AVS SEM and the BLM model) of the bioavailability of contaminants. It could, for example, explain why we are not seeing toxicity in an area where high contaminant concentrations indicate likely toxicity.

In part, EPA's shift in thinking on the use of a separate bioaccumulation test is based on the low levels of bioaccumulative chemicals measured in fish tissue. While mercury, PCBs, and dioxins / furans exceed conservative risk screens for ecological and/or human health risk, most of the levels appear to be within regional or state-wide background ranges. We think the bigger unknown at this point is not bioaccumulation, but direct toxicity to organisms that live or feed in sediment. If we determine through the initial risk assessment calculations that bioaccumulation from sediment up the food chain is a concern, we could add a bioaccumulation study specifically to generate the BSAF values we would need to set sediment cleanup levels."

Discussion: The draft QAPP and the draft Problem Formulation Work Plan submitted by Teck have no mention of collecting tissue from 10-d midge exposures, and this bioaccumulation measure has not yet been explicitly addressed by EPA. What is the status of this test from EPA's point of view? If it will not be required, does the same line of reasoning apply as was explicitly stated above for not requiring *Lumbriculus* tests?

Discussion: Is establishing or estimating invertebrate critical body residues (CBRs) an important analysis goal independent of assessing bioaccumulation in the context of dietary risk to fish? Why or why not? Is there sufficient literature to allow us to interpret invertebrate tissue concentrations from field collected organisms or describe a "consensus" CBR?

Discussion: Is *H. azteca* an acceptable substitute for the *Lumbriculus* in measuring tissue concentrations of bioaccumulated contaminants? Is either species appropriate for <u>all</u> sediment substrates found in the UCR, or would a hybrid approach of *H. azteca* for riverine sediment and *Lumbriculus* for reservoir sediment be useful? Is the body of *H. azteca* residue-response information more advanced than that for *Lumbriculus*?

Discussion: Is it agreed that levels of bioaccumulative chemicals in fish tissue are within regional or state-wide background ranges? How have regional or state-wide background ranges been established?

Agenda: Mussels

Juvenile mussel sediment toxicity tests were outlined in the direction to Teck regarding the Problem Formulation work plan (which was drafted to be consisted with the Sediment LOE). Mussel tests were not included in the draft Problem Formulation work plan submitted by Teck, and the draft Sediment QAPP contained the following:

"Target populations of interest are benthos that live in or on UCR sediment. *H. azteca* and *C. dilutus* have consistently demonstrated to be sensitive indicator organisms for sediment contamination, particularly for metals (Milani et al. 2003); therefore, they are protective of target populations of interest. While it is acknowledged that freshwater mussels (such as the *Anodonta* and *Gonidea* species found in the UCR) may differ from other benthos in their mechanism and duration of exposure to sediment-associated chemicals, an evaluation of sediment bioassay results for the freshwater mussel (*Lampsilis siliquoidae*) relative to the midge (*C. dilutus*) and amphipod (*H. azteca*) supports the use of the standard test organisms. Specifically, any characterization of risk using results with *H. azteca* toxicity tests would be adequately protective for freshwater mussels. Furthermore, there are no approved standard sediment bioassay test methods for freshwater mussels."

DOI says (April 2011; comments on draft Sediment QAPP):

"The statement is made that mussel testing should not be conducted because there is no standard sediment toxicity test described for freshwater mussels. This is not adequate rationale for not testing mussels. Dozens of examples are available regarding the use of non-standard methods for USEPA risk assessments. Even within ongoing UCR risk assessment studies, there are dozens of examples of where methods that have not been standardized are being used (e.g., sturgeon water toxicity testing, sturgeon acute toxicity testing, measurement of SEM and AVS, just to highlight a few)."

DOI says (June 3, 2011; presentation to EPA and PPs):

"ASTM method E2455 for water adapted for conducting sediment toxicity tests with a variety of mussel species (see list of publications)"

EPA says (Aug. 3, 2011; response to Problem Formulation WP suggested revisions):

"We acknowledge that this test was included in the LOE, but we have learned more about this test since then and have come to the conclusion that this test is not ready for use in a regulatory context. There are no commercial labs that can run it and the round-robin data available for standard test species is not available for mussel sediment toxicity tests. Because our understanding of the variability and repeatability of the test is so limited, we would not be comfortable making remedial decisions with the data it would generate. We believe that the other species, combined with mussel tissue data collected in the field will give us sufficient information to support protective cleanup decisions."

EPA says (Sept. 14, 2011; letter to PPs):

"We do not believe that freshwater mussel toxicity tests with sediment are well developed enough to support remedial decisions... We expect the lines of evidence provided by the planned amphipod and midge testing, sediment chemistry, porewater chemistry, invertebrate tissue chemistry and bioavailability models will be sufficient for EPA to make decisions protective of the benthic community."

Discussion: Are juvenile mussels likely to be a more sensitive receptor than *H. azteca* and *C. dilutus*?

Discussion: Should the lack of commercial labs prevent the test from being conducted when government labs are available and have shown the ability to conduct the tests?

Discussion: How do we reconcile the use of non-standard sediment toxicity tests that have already been conducted on this Site in order to assess risk to a receptor of concern (e.g. juvenile sturgeon) with a reluctance to use mussel toxicity tests in order to assess risks to another receptor of concern?